SUPERase•In™

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Cat#	Product Name	Size
2694	SUPERase•In™ (20 U/µl)	2500 U
2696	SUPERase•In™ (20 U/µl)	10,000 U

- **Most effective RNase inhibitor available - inhibits RNase A, B, C, T1 and 1**
- **More powerful than placental ribonuclease inhibitor**
- **Greater protection against RNA degradation**
- **No interference with RNA polymerase, reverse transcriptase, or Taq polymerase**
- **Active up to 65°C and from pH 5.5 to 8.5**
- **No DTT requirement**

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SUPERase•In™ RNase Inhibitor (patent pending) is a protein based inhibitor of nonhuman origin that noncovalently binds and inhibits the most common and troublesome RNases including RNase A, B, C, 1 and T1. SUPERase•In can be used in any application where RNase contamination could be problematic. It is ideal for in vitro transcription and translation, cDNA synthesis, RT-PCR, and preparation of RNase-free antibodies. Because it inhibits a broader range of RNases than traditional RNase inhibitors, SUPERase•In is the most effective RNase inhibitor available providing a higher level of protection against degradation.

The most widely used RNase inhibitor is placental ribonuclease inhibitor (RI), sold as Ribonuclease Inhibitor Protein. SUPERase•In is distinct from RI in that it has a more robust interaction with RNases, has no DTT requirement for activity, and inhibits a

Ribonuclease Inhibitors: Defining a Unit

The traditional unit assay for ribonuclease inhibitors has been the cyclic CMP assay. While this

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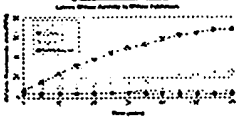
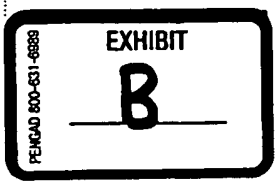


RNase Activity In Ribonuclease Inhibitor Preparations.



broader range of RNases. While placental ribonuclease inhibitor is effective only against RNase A-type enzymes (e.g., RNases A, B, and C), SUPERase•In also inhibits RNase I and T1. SUPERase•In does not interfere with other enzymes such as RNA polymerases, reverse transcriptase or Taq polymerase. Additionally, SUPERase•In is active up to 65° C and over a pH range of 5.5 to 8.5.

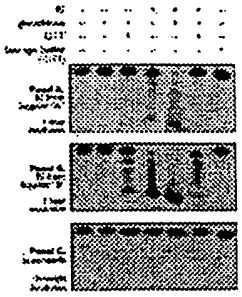
Unit Definition: One unit of SUPERase•In is the amount of protein required to completely block the degradation of 2.5 µg of radiolabeled RNA probe by at least 50 pg of RNase A, 1 U RNase I and 0.15 U RNase T1 as judged by 8 M Urea/5% PAGE and autoradiography.

In practice, 1 U of SUPERase•In defined in this manner will equal or exceed the activity of 1 U of placental ribonuclease inhibitor (RI) defined by the cCMP assay.

assay does demonstrate inhibition of RNase A, it is limited in that it provides only an indirect measure of RNA degradation. Because cCMP is an artificial substrate for RNase, its hydrolysis does not always correlate with actual RNA degradation. In addition, this assay can only be used for RNase A-type ribonucleases and their inhibitors because cCMP is not a substrate for many other ribonucleases.

Ambion has recently developed a new unit assay for ribonuclease inhibitors. It is a functional assay that directly measures the ability of the inhibitor to block RNA degradation. A radiolabeled RNA is exposed to various RNases with and without the inhibitor. The results are analyzed on a polyacrylamide gel to determine the degree to which RNA degradation is inhibited. This functional assay provides direct information about the inhibitor's ability to block RNA degradation.

SUPERase•In is currently the only ribonuclease inhibitor for which the unit activity is defined by such a functional assay.



RNase Activity Measured by Denaturing PAGE and Autoradiography

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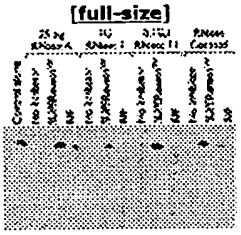

SUPERase•In™ vs. Placental Ribonuclease Inhibitor.

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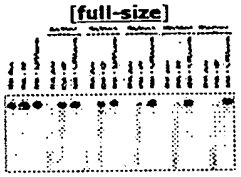

SUPERase•In™ Proves to be a More Robust RNase Inhibitor than RIP.

Placental Ribonuclease Inhibtor (RI)

One of the most commonly used tools to combat RNase contamination is placental ribonuclease inhibitor, a protein that binds RNase A. While originally isolated from placenta, RNase Inhibitor has since been cloned and is sold by Ambion under the generic name of RNase Inhibitor Protein, or RI. RI is often mistaken for a different unique enzyme. While this protein can be useful in solving many RNase contamination problems, its function is often misunderstood. RNase inhibitor proteins do not destroy RNases. The mode of inhibition is noncompetitive; the inhibitor binds RNase A-type enzymes in a 1:1 ratio. To maintain activity, RI requires DTT. Because RI complexes with active RNases, the denaturation or oxidation of the inhibitor can result in the release of active RNases. When using RI, care should be taken to avoid any procedure, such as heat denaturation or addition of SDS, that might result in the release of active RNases.

RELATED PRODUCTS

RNaseZap® RNase Decontamination Solution